

We claim

1. A PNA probe comprising a nucleobase sequence suitable for the analysis of one or more *Staphylococcus* species other than *S. aureus*.
- 5 2. A PNA probe comprising a nucleobase sequence suitable for the analysis of two or more *Staphylococcus* species other than *S. aureus*.
- 10 3. A PNA probe of claims 1-2, wherein a target sequence of the *Staphylococcus* species is rRNA, rDNA or a complement of rRNA or rDNA.
- 15 4. A PNA probe of claim 1-2, comprising a nucleobase sequence suitable for the analysis of *Staphylococcus epidermidis*, said PNA probe being complementary to a target sequence of *Staphylococcus epidermidis* rRNA or rDNA or its complement.
- 20 5. A PNA probe of claim 1-2, comprising a nucleobase sequence suitable for the analysis of *Staphylococcus hominis*, said PNA probe being complementary to a target sequence of *Staphylococcus hominis* rRNA or rDNA or its complement.
- 25 6. A PNA probe of claim 1-2, comprising a nucleobase sequence suitable for the analysis of *Staphylococcus haemolyticus*, said PNA probe being complementary to a target sequence of *Staphylococcus haemolyticus* rRNA or rDNA or its complement.
- 30 7. A PNA probe of claim 1-2, comprising a nucleobase sequence suitable for the analysis of *Staphylococcus lugdunensis*, said PNA probe being complementary to a target sequence of *Staphylococcus lugdunensis* rRNA or rDNA or its complement.
8. A PNA probe of claim 1-2, comprising a nucleobase sequence suitable for the analysis of *Staphylococcus saprophyticus* said PNA probe being

complementary to a target sequence of *Staphylococcus saprophyticus* rRNA or rDNA or its complement.

- 5 9. A PNA probe of claim 1, comprising a nucleobase sequence suitable for the analysis of *Staphylococcus epidermidis* and one or more other *Staphylococcus* species other than *Staphylococcus aureus*, said PNA probe being complementary to a target sequence of *Staphylococcus epidermidis* rRNA or rDNA or its complement
- 10 10. A PNA probe of claim 1, comprising a nucleobase sequence suitable for the analysis of *Staphylococcus hominis* and one or more other *Staphylococcus* species other than *Staphylococcus aureus*, said PNA probe being complementary to a target sequence of *Staphylococcus hominis* rRNA or rDNA or its complement.
- 15 11. A PNA probe of claim 1, comprising a nucleobase sequence suitable for the analysis of *Staphylococcus haemolyticus* and one or more other *Staphylococcus* species other than *Staphylococcus aureus*, said PNA probe being complementary to a target sequence of *Staphylococcus haemolyticus* rRNA or rDNA or its complement.
- 20 12. A PNA probe of claim 1, comprising a nucleobase sequence suitable for the analysis of *Staphylococcus lugdunensis* and one or more other *Staphylococcus* species other than *Staphylococcus aureus*, said PNA probe being complementary to a target sequence of *Staphylococcus lugdunensis* rRNA or rDNA or its complement.
- 25 13. A PNA probe of claim 1, comprising a nucleobase sequence suitable for the analysis of *Staphylococcus saprophyticus* and one or more other *Staphylococcus* species other than *Staphylococcus aureus*, said PNA probe being complementary to a target sequence of *Staphylococcus saprophyticus* rRNA or rDNA or its complement.
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14. The PNA probe of claims 1, 2, or 9-13, wherein at least a portion of the probe is at least about 86% identical to the nucleobase sequence or complement thereof selected from the following sequences: TCT-AAC-ATG-TTC-TTT (Seq. Id. No. 1), TCT-AGT-CTG-TTC-TTT (Seq. Id. No. 2), TCT-AAT-ATA-TTC-CTT (Seq. Id. No. 3), TCT-AAT-ATA-TAC-TTT (Seq. Id. No. 4), GCT-CCA-AAT-GGT-TAC (Seq. Id. No. 5), TCC-TCG-TCT-GTT-CGC (Seq. Id. No. 6), CTC-CTT-ATC-TGT-TCG-C (Seq. Id. No. 7), and CTC-CTT-GTC-TGT-TCG-C (Seq. Id. No. 8), CTT-CTC-ATC-TGT-TCG-C (Seq. Id. No. 9), TCC-TCG-TCC-GTT-CGC (Seq. Id. No. 10), TCC-TTG-TCC-GTT-CGC (Seq. Id. No. 11).
15. The PNA probe of claims 1, 2 or 9-13, wherein at least a portion of the probe is selected from the following sequences: TCT-AAC-ATG-TTC-TTT (Seq. Id. No. 1), TCT-AGT-CTG-TTC-TTT (Seq. Id. No. 2), TCT-AAT-ATA-TTC-CTT (Seq. Id. No. 3), TCT-AAT-ATA-TAC-TTT (Seq. Id. No. 4), GCT-CCA-AAT-GGT-TAC (Seq. Id. No. 5), TCC-TCG-TGT-GTT-CGC (Seq. Id. No. 6), CTC-CTT-ATC-TGT-TCG-C (Seq. Id. No. 7), and CTC-CTT-GTC-TGT-TCG-C (Seq. Id. No. 8), CTT-CTC-ATC-TGT-TCG-C (Seq. Id. No. 9), TCC-TCG-TCC-GTT-CGC (Seq. Id. No. 10), TCC-TTG-TCC-GTT-CGC (Seq. Id. No. 11).
16. The PNA probe of claims 1, 2, or 9-13, wherein the probe sequence is 8-17 subunits in length.
17. The PNA probe of claims 1, 2, or 9-13, wherein the probe is labeled with at least one detectable moiety.
18. The PNA probe of claim 17, wherein the detectable moiety or moieties are selected from the group consisting of: a conjugate, a branched detection system, a chromophore, a fluorophore, a spin label, a radioisotope, an enzyme, a hapten, an acridinium ester and a luminescent compound.
19. The PNA probe of claim 17, wherein the probe is self-reporting.

20. The PNA probe of claims 19, wherein the probe is a PNA Linear Beacon.
- 5 21. The PNA probe of claims 1, 2, or 9-13, wherein the probe is unlabeled.
22. The PNA probe of claim 21, wherein the probe is bound to a support.
- 10 23. The PNA probe of claims 1, 2, or 9-13, wherein the probe further comprises a spacer or a linker.
24. The PNA probe of claims 1, 2, or 9-13, wherein *in situ* hybridization is used for analysis of one or more *Staphylococcus* species other than *S. aureus*.
- 15 25. A PNA probe set comprising, one or more PNA probes of claims 1-24 and at least one PNA probe for the analysis of *S. aureus*.
- 20 26. A PNA probe set of claim 25, wherein the probes are differently labeled for independent analysis of two or more *Staphylococcus* species.
- 25 27. A PNA probe set of claim 25, wherein the PNA probes comprise PNA probes wherein at least a portion of each probe is selected from the following sequences: TCC-TCG-TCT-GTT-CGC (Seq. Id. No. 6), CTC-CTT-ATC-TGT-TCG-C (Seq. Id. No. 7), CTC-CTT-GTC-TGT-TCG-C (Seq. Id. No. 8), CTT-CTC-ATC-TGT-TCG-C (Seq. Id. No. 9), TCC-TCG-TCC-GTT-CGC (Seq. Id. No. 10), TCC-TTG-TCC-GTT-CGC (Seq. Id. No. 11).
- 30 28. A PNA probe set of claim 25, wherein the at least one PNA probe for the analysis of *S. aureus* is differently labeled from the one or more PNA probes of claims 1-24.

29. A PNA probe set of claim 28, wherein at least a portion of one of the at least one PNA for the analysis of *S. aureus* is selected from the following sequence: GCT-TCT-CGT-CCG-TTC.
- 5 30. A PNA probe set comprising one or more PNA probes of claims 1-24 and at least one PNA blocking probe for the analysis of one or more *Staphylococcus* species other than *S. aureus*.
- 10 31. A PNA probe set of claim 30, further comprising a PNA blocking probe for analysis of *S. aureus*.
- 15 32. A PNA probe set of claim 31, wherein at least a portion of the PNA blocking probe for analysis of *S. aureus* is selected from the following sequences: TCC-TCG-TCT-GTT-CGC (Seq. Id. No. 6), CTC-CTT-ATC-TGT-TCG-C (Seq. Id. No. 7), CTC-CTT-GTC-TGT-TCG-C (Seq. Id. No. 8), CTT-CTC-ATC-TGT-TCG-C (Seq. Id. No. 9), TCC-TCG-TCC-GTT-CGC (Seq. Id. No. 10), TCC-TTG-TCC-GTT-CGC (Seq. Id. No. 11).
- 20 33. A method for the analysis of *Staphylococcus* species other than *S. aureus* in a sample, comprising: a) contacting at least one of the PNA probes of claims 1-24 to the sample, b) hybridizing the PNA probe to a target sequence of *Staphylococcus* species other than *S. aureus* in the sample; and
- 25 c) detecting the hybridization, wherein the detection of hybridization is indicative of the presence, identity and/or amount of *Staphylococcus* species other than *S. aureus* in the sample. ,
- 30 34. A method for the analysis of two or more *Staphylococcus* species, said method comprising: a) contacting a PNA probe set of claims 1-24 to the sample, b) hybridizing the PNA probes to a target sequence of *Staphylococcus* species in the sample; and c) detecting the hybridization, wherein the detection of hybridization is

indicative of the presence, identity and/or amount of *Staphylococcus* species in the sample.

5 35. A method of claims 33 - 34, wherein two or more *Staphylococcus* species are *S. aureus* and one or more *Staphylococcus* species other than *S. aureus*.

36. The method of claims 33-35 for analysis of a cohort of *Staphylococcus* species other than *S. aureus*.

10 37. The method of claims 36 in which the cohort of *Staphylococcus* species other than *S. aureus* detected is Coagulase-Negative Staphylococci, clinically significant Coagulase-Negative Staphylococci, or a subset of clinically significant Coagulase-Negative Staphylococci.

15 38. The method of claim 33-37, wherein the probes are independently detectable non-independently detectable, or a combination of independently and non-independently detectable, wherein the probes differ from one another by as little as a single base, and are
20 complementary or substantially complementary to partially conserved target regions of phylogenetically related organisms.

25 39. The method of claim 38, wherein probes or probe sets are used to eliminate or reduce cross hybridization between a *Staphylococcus aureus* specific probe, and a *Staphylococcus scheiferi* target.

40. A method of claim 33-39, wherein *S. aureus* and one or more *Staphylococcus* species other than *S. aureus* are simultaneously and independently detected.

30 41. A method according to claim 33-40, wherein the analysis takes place *in situ*.

42. A method according to claim 41, wherein the analysis takes place by fluorescence *in situ* hybridization.

43. The method of claim 33-40, wherein the method is used to detect a nucleic acid comprising a target sequence wherein said nucleic acid has been synthesized or amplified in a reaction.

44. The method of claim 43, wherein preferred nucleic acid synthesis or nucleic acid amplification reactions are selected from the group consisting of: Polymerase Chain Reaction (PCR), Ligase Chain Reaction (LCR), Strand Displacement Amplification (SDA), Transcription-Mediated Amplification (TMA), Rolling Circle Amplification (RCA) and Q beta replicase.

45. The method of claim 33-44, wherein the method further comprises adding at least one blocking probe to reduce or eliminate hybridization of the PNA probe to non-target sequence.

46. The method of claim 33-40, wherein the target sequence is immobilized to a surface.

47. The method of claim 33-40, wherein said PNA probe is immobilized to a surface.

48. The method of claim 33-40, wherein said PNA probe is one component of an array.

49. The method of claims 33-48, wherein the sample is a biological sample.

50. The method of claim 49, wherein the biological sample is blood, urine, secretion, sweat, sputum, stool, mucous, or cultures thereof.

51. A kit suitable for performing an assay for analysis of one or more *Staphylococcus* species other than *S. aureus* in a sample, wherein said kit comprises: a) a PNA probe according to claim 1 to 24 and b) other reagents or compositions necessary to perform the assay.

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52. The kit of claim 51, wherein one or more *Staphylococcus* species other than *S. aureus* and at least one other microorganism optionally present in a sample are independently detected, identified and/or quantitated.

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53. The kit of claim 52, wherein one or more *Staphylococcus* species other than *S. aureus* and *S. aureus* optionally present in a sample are independently detected, identified and/or quantitated.

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54. The kit of claim 51-53, wherein one or more *Staphylococcus* species other than *S. aureus* optionally present in a sample is detected, identified and/or quantitated and its susceptibility to antimicrobial agents is determined.

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55. The kit of claim 51-54, wherein one or more *Staphylococcus* species other than *S. aureus* detected is Coagulase-Negative Staphylococci, clinically significant Coagulase-Negative Staphylococci, or a subset of clinically significant Coagulase-Negative Staphylococci.

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56. The kit of claim 51-55, wherein the kit is used in an *in situ* hybridization assay.

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57. The kit of claim 56, wherein the kit is a fluorescence *in situ* hybridization assay for simultaneous, independent (multiplex) identification of one more *Staphylococcus* species other than *S. aureus* and of *S. aureus*.

58. The kit of claim 51-55, wherein the kit is used for a real-time PCR assay.

59. The kit of claim 51-58, wherein the kit is used to examine clinical samples such as clinical specimens or cultures thereof.